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SOME VARIATION OF TERPENES IN MONTANA LODGEPOLE PINE

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ABSTRACT

Intraspecific variation was found in the terpene fraction of oleoresin of Pinus contorta *near Bozeman, Montana. There was a tendency towards* P. banksiana-*type terpenes in 3 trees out of 11 sampled. In these trees α-pinene or β-pinene was the predominant terpene. The "typical" lodgepole pine terpene--β-phellandrene--varied from 8 to 83 percent among the 11 trees.*

Certain extraneous substances can be useful in understanding the origin, development, and relationships of species within a genus, and Mirov (1967) has been a leader in studying these relationships for the turpentine composition of the genus *Pinus*. However, sampling has been inadequate as he readily admitted. Population studies are needed to define "typical" compositions for the various species, subspecies, and varieties involved. Inadequate sampling can give an oversimplified picture of the chemical constituents of a taxon.

According to traditional concepts (Mirov 1948, 1961; Smith 1964, 1967b; Williams and Bannister 1962), the terpene fraction of lodgepole pine (*Pinus contorta* Dougl.) consists primarily of the monocyclic terpene, β-phellandrene. Smith had the advantage of using the gas chromatograph, therefore his work was more precise than the earlier work. Mirov, in his analysis, used steam distillation and the chemical procedures that were conventional at that time. However, there was general agreement in that most of the terpene composition for lodgepole pine was β-phellandrene. Other terpenes, such as

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α-pinene, Δ_3-carene, β-pinene, myrcene, and camphene, were usually never over 10 percent for any single component. Williams and Bannister (1962) had similar results for lodgepole pine grown in New Zealand.

Mirov (1961) reported that the turpentine of jack pine (*P. banksiana* Lamb.) is composed of mostly α-pinene and β-pinene. Further, he had reported earlier (Mirov 1956) that natural hybrids of *P. contorta* and *P. banksiana* tended to be intermediate in terpene chemistry. Smith (1967a) reported similar intermediate traits in artificial hybrids in California.

More recently, Zavarin, Critchfield, and Snajberk (1969), in comparing turpentine composition of *P. contorta* X *banksiana* hybrids with that of jack pine and lodgepole pine have shown evidence of possible introgression[2] of jack pine into lodgepole pine. Rocky Mountain lodgepole pine apparently has a tendency to deviate from "lodgepole pine" towards "jack pine" in turpentine composition.

Supporting evidence of this introgression can also be found in older studies. Mirov (1956) found that individual lodgepole pines, 30 to 50 miles outside the range of jack pine, possessed a range of physical properties of turpentine samples characteristic of both lodgepole pine and jack pine. Critchfield (1957) proposed the possibility of introgression in lodgepole pine based upon "jack pine-type" cone orientation in the northeastern part of the range of lodgepole pine.

We sampled 11 lodgepole pine trees in the northern Rocky Mountains in Montana using a technique similar to Smith's (1961) closed-face microtap. The samples were analyzed by gas chromatography and, in support of Zavarin, Critchfield, and Snajberk (1969), we found the terpene composition had a tendency to deviate from "typical" lodgepole pine among a relatively small number of trees. The percentage of β-phellandrene in the terpene fraction varied considerably more than that usually reported for *P. contorta*. When the β-phellandrene percentage was low, either α-pinene, β-pinene, or Δ_3-carene was high.

EXPERIMENTAL

Oleoresin was collected from the tree bole 4.5 feet above the ground with closed-face microtaps from 11 dominant or codominant lodgepole pine (*P. contorta* var. *latifolia* Engelm.) in Hyalite Canyon 10 miles south of Bozeman, Montana. Some of the trees were young and vigorous: trees 1-5 and 11 (20-38 years old, 3-6 inches d.b.h., and 16-46 feet tall). The others were mature or overmature: trees 6-10 (73-171 years old, 10-19″ d.b.h., and 45-90′ tall).

The freshly collected oleoresin was stored under an inert atmosphere at -5° C. The terpene analyses were run on two columns in order to separate all of the components:

(1) a 15-foot X 3/16-inch 10 percent Versamid 900 column at 115° C., and

(2) a 12-foot X 3/16-inch 20 percent Carbowax 20M column at 115° C.

Conditions for operation of the F and M Model 700 gas chromatograph were:

Injector temperature.	300° C.
Column temperature.	115° C.
Detector temperature.	300° C.
Helium flow	150 ml./min.
Sample size	0.5-3.0 μ liter
Solid support	60/80 mesh Diatoport S.

[2]The spread of genes away from an area of natural hybridization.

Table 1.--*Terpene composition of lodgepole pine oleoresin from Montana, 1970.*

Tree number	α-pinene	Camphene	Myrcene	β-pinene	Δ_3-carene	α-terpinene	Limonene	β-phellandrene	p-cymene	Allo-ocimene
	Percent									
1	[1]2	1	3	13	57*	4	1	8	4	9
2	9	[2]Tr	5	14	10	-	3	59*	Tr	-
3	4	1	4	8	17	-	-	64*	3	-
4	10	2	1	58*	7	-	2	20	2	-
5	7	Tr	3	28	27	-	2	32*	Tr	2
6	5	1	4	13	25	-	1	49*	2	-
7	2	Tr	3	7	3	2	1	83*	2	2
8	50*	Tr	3	6	27	Tr	1	11	Tr	-
9	5	Tr	1	13	10	Tr	2	65*	7	-
10	6	Tr	3	32	2	Tr	2	54*	-	-
11	9	Tr	2	52*	6	-	1	23	-	-
Average	10	Tr	3	22	17	Tr	1	42	2	1
Range	2-50	Tr-2	1-5	6-58	2-57	0-4	0-3	8-83	0-4	0-9

[1]The combined, corrected, or averaged values from both columns used.
[2]Trace values less than 1 percent.
* Major constituent.

Myrcene and Δ_3-carene do not separate on the Carbowax column so these were determined on the Versamid column. The Carbowax column was better than the Versamid column in separating β-pinene. Thus, both columns were used and the results were combined, corrected, and averaged.

RESULTS AND DISCUSSION

The major terpene constitutents of the trees sampled were β-phellandrene, Δ_3-carene, α-pinene, and β-pinene (table 1). The percentage of β-phellandrene (the "typical" lodgepole pine terpene) varied from 8 to 83 percent. In fact, for these samples, β-phellandrene was the major constituent in only 7 trees out of the 11. For the remainder, in 2 trees, the largest peak was β-pinene. In one it was α-pinene. In one it was Δ_3-carene. Other terpenes found in lesser amounts were camphene, myrcene, α-terpinene, limonene, p-cymene, and allo-ocimene; generally, these were on the order of a few percent when present.

This variation in terpene constituents, especially in trees 4, 8, and 11, supports the hypothesis of Zavarin, Critchfield, and Snajberk (1969), that the terpenes of lodgepole pine have been genetically influenced by jack pine at some distance from the zone of overlap. The range in percentage of β-phellandrene is considerable, and usually when β-phellandrene is low there is a corresponding rise in either Δ_3-carene, α-pinene, or β-pinene.

There is a difference in the way the data are presented here, and in the paper by Zavarin, Critchfield, and Snajberk (1969). These data are presented as a percent of the terpene fraction as compared to Zavarin's percent of total resin. However, one can readily see that here in the Montana trees we have α-pinene and β-pinene (the "jack pine" terpenes) showing up strongly in a lodgepole pine population some 600 miles from the nearest jack pine.

Zavarin, Critchfield, and Snajberk (1969) made an excellent review of current thinking on natural hybridization and introgression of these two interesting species. Apparently, our data support their thinking.

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